Chemistry and Physics of Lipids 221 (2019) 15–23

Contents lists available at ScienceDirect

Chemistry and Physics of Lipids

journal homepage: www.elsevier.com/locate/chemphyslip

# Comprehensive analysis of the phospholipids and phytosterols in *Schisandra chinensis* oil by UPLC-Q/TOF- $MS^E$

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ARTICLE INFO

*Keywords:*
*S. chinensis* oil
Hipposterol
Theonellasterol
Conicasterol
Penasterol

ABSTRACT

Characterization of phospholipids (PLs) and phytosterols were determined in *Schisandra chinensis* (*S. chinensis*) oil by UPLC-Q/TOF-$MS^E$. The determination process was based on feature fragment information of components generated by the $MS^E$ detector. A total of 49 and 39 PLs were identified in *S. chinensis* oil under negative and positive ion mode, respectively. The classes of PLs included phosphatidic acids (PAs), phosphatidylethanolamines (PEs), phosphatidylglycerols (PGs), phosphatidylinositols (PIs), phosphatidylserines (PSs) and phosphatidylcholines (PCs). The most diverse species of PLs detected were PIs and PCs, accounting for 12 and 14, respectively. The analysis of quantification indicated that PEs and PCs were the most abundant constituents in *S. chinensis* oil, accounting for 122.85 ± 3.82 and 85.61 ± 2.12 μg/g, respectively. Besides, thirteen kinds of phytosterols were tentatively identified in *S. chinensis* oil under positive ion mode, among which, conicasterol C was the most abundant component, accounting for 22.02 ± 0.98 μg/g. Brassicasterol, campesterol, secosterol-B and herbasterol were also determined in *S. chinensis* oil. These results might be meaningful in the quality assessment and function evaluation of *S. chinensis* oil.

## 1. Introduction

Plants oils are mainly composed of triglycerides and small amounts of complex minor substances (2–5%) (Cañabate-Díaz et al., 2007), such as free fatty acids, phospholipids (PLs), phytosterols, pigments and vitamins. These minor compounds play an important role in the discrimination, function and quality evaluation of oil species. *Schisandra chinensis* (*S. chinensis*) is a traditional Chinese herbal plant with many pharmaceutical activities (Sowndhararajan et al., 2018). It has been authenticated as a new homologous product of medicine and food (Mocan et al., 2016). It is reported that *S. chinensis* contains about 10–50% of oils (dry weight) (Panossian and Wikman, 2008; Mocan et al., 2016), yet there are few studies which exploit the minor lipid components of *S. chinensis* oil.

PLs, one of the minor lipid components in plants oils, are a highly abundant and diverse collection of biologically relevant lipids and structural lipids (Stępniewska et al., 2017; Ali et al., 2017). In addition, PLs are a major constituent of the cell membranes, which might influence the function of the cell under some alterations (Taguchi et al., 2000). Moreover, the latest research showed that PLs are a good source of arachidonic acid, which is important in metabolism, especially in the synthesis of prostaglandins and leukotrienes (Furse and De Kroon, 2015). Generally, PLs own higher bioactivity in comparison to triglycerides lipids. The investigation of PLs in *S. chinensis* oil might be helpful for us to make a better recognition of its nutritional values.

The analysis of PLs is a challenge due to their complex chemical structures and various acyl chains. Up to now, the analysis methods of PLs have developed from the conventional thin layer chromatography (TLC) techniques to more progressive mass spectrometry technologies (Henderson et al., 2011), and mass spectrometry has been extensively used in the analysis of lipids (Murphy et al., 2001; Song et al., 2017). However, due to the ion suppression, the simultaneous occurrence of structural and positional isomers and low abundance of PLs fractions, the rapid identification of PLs is still being explored (Zhou et al., 2018; Liang et al., 2018). For example, PLs can be identified in both two ion modes (Murphy et al., 2001), while some researchers identified them only in the negative ion mode (Song et al., 2017). In addition, most of the research recognized the PLs based on the reference standard materials (Jones et al., 2015; Ali et al., 2017). Moreover, it is reported that the length of the acyl chain and its unsaturation have a significant effect on instrument response for every phospholipid species. Nevertheless, quantitative compositional data of PLs can be obtained with ESI-MS (Koivusalo et al., 2001).

Aside from PLs, phytosterols are natural and minor components

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https://doi.org/10.1016/j.chemphyslip.2019.03.003
Received 29 December 2018; Received in revised form 26 January 2019; Accepted 6 March 2019
Available online 07 March 2019
0009-3084/ 

**Table 1**
Characteristic fragments of different PLs classes in UPLC-Q/TOF-MS$^E$.

| PL type | Precursor | | Characteristic fragment | | Fatty acyl identification fragments | |
|---|---|---|---|---|---|---|
| | ESI$^+$ | ESI$^-$ | ESI$^+$ | ESI$^-$ | ESI$^+$ | ESI$^-$ |
| PC | $[M+Na]^+$, $[M+NH_4]^+$ | $[M-H^-$ | 184.0732 | | $[M+H]^+$ | $[M-H]^-$ |
| PA | $[M+Na]^+$, $[M+NH_4]^+$ | $[M-H]^-$ | – | 152.9953 (PL1), 78.9585 (PL2), 171.0059 | $[M+H]^+$ | $[M-H]^-$ |
| PE | $[M+Na]^+$, $[M+NH_4]^+$ | $[M-H]^-$ | 142.0269 | 140.0113, 78.9585, 171.0059 | $[M+H]^+$ | $[M-H]^-$ |
| PS | $[M+Na]^+$, $[M+NH_4]^+$ | $[M-H]^-$ | 88.0399, 226.0481 | 224.0324, 152.9953, 171.0059, 78.9585 | $[M+H]^+$ | $[M-H]^-$ |
| PG | $[M+Na]^+$, $[M+NH_4]^+$ | $[M-H]^-$ | – | 227.0321, 152.9953, 171.0059, 78.9585 | $[M+H]^+$ | $[M-H]^-$ |
| PI | $[M+Na]^+$, $[M+NH_4]^+$ | $[M-H]^-$ | 261.0735 | 259.0219, 241.0113, 299.0532, 152.9953, 78.9585 | $[M+H]^+$ | $[M-H]^-$ |

existing in oils derived from various plant (Szterk and Pakuła, 2016). Over the past few years, phytosterols have attracted considerable interest because of their alleged ability to reduce blood cholesterol levels and lower the risk of cardiovascular disease (Chen et al., 2017). Recently, the composition of phytosterols in olives has been widely applied in the determination of oil adulteration or authenticity (Lerma-García et al., 2011). In addition, the content of phytosterols in oils has been used to indicate the maturity of oil extraction materials, extraction technique, storage conditions and oil quality (Broughton et al., 2018). Phytosterols together with other secondary metabolites have also been recognized as cancer preventive biologically active substances (Alberici et al., 2016).

Likewise, for the analysis of phytosterols, GC-FID is the standard technique. It has been widely applied in the study of phytosterols. However, it has many disadvantages, such as time-consuming, laborious and reagent-dissipating (Chen et al., 2017). With the development of analytical technique, phytosterols have been investigated by various novel approaches. The methods include high-performance liquid chromatography (HPLC), liquid chromatography-atmospheric pressure chemical ionization-mass spectrometry (LC-APCI-MS), and several direct MS protocols (electrospray ionization, desorption electrospray ionization and direct analysis in real time) (Gachumi and El-Aneed, 2017; Novak et al., 2018).

Generally, the current mass spectrometry strategies for components characterization rely heavily on the use of gas-phase dissociation techniques to fragment intact precursor ions to yield structure specific product ions. Therefore, the component structures can be determined by their fragmentation. Recently, UPLC-Q/TOF-MS$^E$ has been widely applied in complicated component analysis because of its rapid and sensitive separation, and the precision of mass measurement (Donazzolo et al., 2017). Consequently, the problems linked with multiple constituent separation and characterization could be solved. The identification could be achieved in a single analytical process (Noguer et al., 2017; Liang et al., 2018).

In this study, we have explored the applicability of ESI coupled with MS$^E$ data acquisition process to characterize PLs and phytosterols in *S. chinensis* oil. The method permitted the separation of PLs and phytosterols without tedious pretreatment. All the analysis process was completed in 17 min. The results of this study would be significantly useful for the nutrition investigation and dietary application of *S. chinensis* oil.

## 2. Materials and methods

### 2.1. Samples

*S. chinensis* fruits were collected from Lan Xinbao Co., Ltd (Jilin, China). They were determined by food and drug administration of Yanbian (Jilin, China). As a result, they belonged to the wild type of north *S. chinensis* fruits. The *S. chinensis* oil was extracted by ultrasonication-assisted n-hexane extraction (UAE) (Xinzhi Biotechnology Co., Ltd, KQ3200E, Ningbo, China). The UAE process was performed at an ultrasonic output power of 240 W and a frequency of 40 kHz. The sonication time and temperature were 30 min and 20 °C, respectively. Then, the mixtures were left for 3–4 h, and the supernatant liquid was collected and concentrated at 45 °C using a rotary evaporator (Eyela, N-1100, Shanghai, China) to obtain the oils. All the oils were stored at 4 °C until use.

### 2.2. Standards and reagents

The PLs standard of 1,2-(9Z,12Z-octadecadienoyl)-*sn-glycero*-3-phosphocholine [PC (18:2/18:2)] (PC (36:4), > 98%) and stigmasterol (> 99%) were obtained from ANPEL Laboratory Technologies Inc (Shanghai, China). Formic acid, n-hexane and chloroform were purchased from the Sinopharm Chemical Reagent Co. Ltd (Shanghai, China). The solvents used for separation and chromatographic analysis were supplied by Fisher Scientific Co. Ltd (Shanghai, China), including methanol and acetonitrile (HPLC grade). Water was prepared by a Milli-Q water purification system (Millipore Co. Ltd, Milford, USA). The protective gases were nitrogen (99.999%, Shanghai Likang Co. Ltd, Shanghai, China) and helium (99.999%, Shanghai Likang Co. Ltd, Shanghai, China).

### 2.3. Analysis of the PLs and phytosterols of S. chinensis oil by UPLC-Q/TOF-MS$^E$ system

#### 2.3.1. UPLC conditions

The aliquots of 0.10 g oil sample were re-dissolved in 10 mL chloroform/methanol ($v/v$ = 1:3) for further PL and phytosterol analysis, respectively. The separation of oils was performed on a Waters I-Class Acquity UPLC (Waters, Shanghai, China) system with BHC C18 column (100 mm × 2.1 mm, 1.7 μm). The eluent A was 0.1% formic acid water and eluent B was 0.1% formic acid/methanol/acetonitrile. The elution was operated at a flow rate of 0.4 mL/min. The column temperature was 50 °C. The injection volume was 1 μL. Sample separation was operated under the following conditions: 0 - 0.5 min, 90% A linear and 10% B linear; 0.5–2 min, 50% A and 50% B; 2–7 min, 20% A linear and 80% B linear, 7–15 min: 100% B; 15–17 min, 99% A and 10% B. A blank sample (methanol) was added in the sequence run before and after each group of samples for column cleaning. All the samples were analyzed in triplicate.

#### 2.3.2. Mass spectrometric analysis

Mass spectrometric data were acquired on the VION-IMS-Q-TOF mass spectrometer (Waters, Shanghai, China) using both positive and negative ion electrospray ionization. The MS data were acquired in the range of 50–1000 *m/z*. The scan time for each function was set as 0.2 s. Ion monitoring conditions were defined as capillary voltage 2.0 kV, source temperature 120 °C, and desolvation temperature 500 °C. The condition of two functions in MS$^E$ experiments were as follows: function one was acted as parent ions detection mode (MS$^E$-L), with a collision energy of 6 V; function two was played as fragments ion detection mode (MS$^E$-H), with a collision energy ramp of 20–40 V. The switching frequency from function one to function two was 30 Hz. Nitrogen (99.999%) was used as the desolvation gas (1000 L/h) and cone gas (50 L/h). Data were analyzed and processed using UNIFI 1.8.1

**Table 2**
Identified PLs from *S. chinensis* oil in the negative ion mode.

| Head | Species | RT/min | Neutral mass | Adducts | Formula | Mass error (ppm) | $MS^E$-H: Feature fragments | $MS^E$-H: Fatty acyl chains | Degree | Semi-quantitive content (μg/g) |
|---|---|---|---|---|---|---|---|---|---|---|
| PA/ESI$^-$ | | | | | | | | | | |
| | 24:3 | 3.94 | 530.3009 | -H | $C_{27}H_{47}O_8P$ | 2.8 | 78.9591 (PL1) | ND | B | 3.75 ± 0.21 |
| | 27:4 | 5.50 | 570.3322 | -H | $C_{30}H_{51}O_8P$ | 3.3 | 152.9958 (PL2) | ND | B | 3.77 ± 0.18 |
| | 17:0 | 6.34 | 424.2590 | -H | $C_{20}H_{41}O_7P$ | 1.6 | 171.0064 (PA), 152.9958 (PL2) | ND | B | 3.37 ± 0.24 |
| | 19:2 | 7.38 | 448.2590 | -H | $C_{22}H_{41}O_7P$ | 1.3 | 171.0064 (PG/PA) | ND | C | 3.24 ± 0.23 |
| | 33:2 | 7.48 | 658.4574 | -H | $C_{36}H_{67}O_8P$ | −1.6 | 171.0064 (PG/PA) | ND | B | 3.22 ± 0.18 |
| | 26:3 | 8.91 | 558.3322 | -H | $C_{29}H_{51}O_8P$ | 3.1 | ND | C (18:2) C (8:1) | B | 4.30 ± 0.32 |
| | 39:5 | 9.50 | 736.5043 | -H | $C_{42}H_{73}O_8P$ | −2.9 | 171.0064 (PG/PA) | ND | B | 5.92 ± 0.41 |
| | 37:4 | 9.51 | 710.4887 | -H | $C_{40}H_{71}O_8P$ | 0.1 | 171.0064 (PG/PA) | ND | B | 4.09 ± 0.34 |
| PE/ESI$^-$ | | | | | | | | | | |
| | 20:4 | 5.78 | 501.2855 | -H | $C_{25}H_{44}NO_7P$ | −0.5 | ND | ND | C | 3.05 ± 0.21 |
| | 18:3 | 6.60 | 475.2699 | -H | $C_{23}H_{42}NO_7P$ | −2.3 | ND | C (18:3) | B | 9.60 ± 0.14 |
| | 26:5 | 7.21 | 597.3431 | -H | $C_{31}H_{52}NO_8P$ | −1.5 | 78.9591 (PL1) | ND | B | 3.75 ± 0.21 |
| | 18:2 | 7.29 | 477.2855 | -H | $C_{23}H_{44}NO_7P$ | −1.7 | 140.0118 (PE), 78.9591 (PL1) | C (18:2) | A | 61.44 ± 2.64 |
| | 16:0 | 7.84 | 453.2855 | -H | $C_{21}H_{44}NO_7P$ | −1.0 | ND | C (16:0) | B | 18.96 ± 0.43 |
| | 20:0 | 8.60 | 509.3481 | -H | $C_{25}H_{52}NO_7P$ | 0.6 | ND | ND | C | 3.13 ± 0.25 |
| | 36:4 | 10.04 | 739.5152 | -H | $C_{41}H_{74}NO_8P$ | 3.1 | 140.0118 (PE) | C (18:2) C (18:2) | A | 35.25 ± 1.22 |
| | 34:2 | 10.26 | 715.5152 | -H | $C_{39}H_{74}NO_8P$ | −1.6 | 140.0118 (PE), 78.9591 (PL1), | C (16:0) C (18:2) | A | 26.16 ± 0.89 |
| | 33:0 | 10.59 | 705.5309 | -H | $C_{38}H_{76}NO_8P$ | −1.1 | ND | ND | C | 3.16 ± 0.23 |
| | 41:0 | 10.64 | 817.6561 | -H | $C_{46}H_{92}NO_8P$ | 3.0 | ND | C (20:0) C (21:0) | B | 3.17 ± 0.33 |
| PG/ESI$^-$ | | | | | | | | | | |
| | 18:2 | 6.78 | 508.2801 | -H | $C_{24}H_{45}O_9P$ | 3.6 | 78.9591 (PL1), 152.9958 (PL2) | C (18:2) | A | 3.35 ± 0.25 |
| | 31:3 | 7.05 | 702.4472 | -H | $C_{37}H_{67}O_{10}P$ | 3.1 | 152.9958 (PL2) | ND | B | 3.77 ± 0.24 |
| | 34:3 | 7.31 | 744.4941 | -H | $C_{40}H_{73}O_{10}P$ | 2.0 | 78.9591 (PL1) | ND | B | 4.12 ± 0.37 |
| | 16:0 | 7.55 | 484.2801 | -H | $C_{22}H_{45}O_9P$ | 1.7 | 152.9958 (PL2) | C (16:0) | A | 6.28 ± 0.42 |
| | 18:1 | 7.73 | 510.2958 | -H | $C_{24}H_{47}O_9P$ | −1.9 | ND | C (18:1) | B | 3.87 ± 0.12 |
| | 39:6 | 8.91 | 808.5254 | -H | $C_{45}H_{77}O_{10}P$ | −0.9 | 152.9958 (PL2) | ND | B | 3.57 ± 0.14 |
| | 39:1 | 9.51 | 818.6037 | -H | $C_{45}H_{87}O_{10}P$ | 2.8 | 171.0064 (PG/PA) | ND | B | 4.18 ± 0.21 |
| | 38:3 | 10.29 | 800.5567 | -H | $C_{44}H_{81}O_{10}P$ | −3.0 | 78.9591 (PL1), 152.9958 (PL2) | ND | B | 3.75 ± 0.2 |
| | 41:0 | 10.64 | 848.6506 | -H | $C_{47}H_{93}O_{10}P$ | −0.3 | 171.0064 (PG/PA), 152.9958 (PL2) | C (20:0) C (21:0) | A | 6.22 ± 0.44 |
| PI/ESI$^-$ | | | | | | | | | | |
| | 19:0 | 3.99 | 614.3431 | -H | $C_{28}H_{55}O_{12}P$ | −1.1 | 259.0224 (PI), 78.9591(PL1) | ND | B | 3.78 ± 0.24 |
| | 20:0 | 5.34 | 628.3588 | -H | $C_{29}H_{57}O_{12}P$ | −1.8 | 152.9958 (PL2) | ND | B | 4.15 ± 0.24 |
| | 18:3 | 5.85 | 594.2805 | -H | $C_{27}H_{47}O_{12}P$ | 2.6 | ND | C (18:3) | B | 3.13 ± 0.43 |
| | 33:5 | 6.17 | 814.4632 | -H | $C_{42}H_{71}O_{13}P$ | −0.9 | ND | ND | C | 3.13 ± 0.37 |
| | 36:6 | 6.34 | 854.4945 | -H | $C_{45}H_{75}O_{13}P$ | 2.6 | 152.9958 (PL2) | C (18:2) C (18:4) | A | 3.51 ± 0.23 |
| | 31:2 | 6.35 | 792.4789 | -H | $C_{40}H_{73}O_{13}P$ | −2.1 | 152.9958 (PL2) | C (14:0) C (17:2) | A | 3.55 ± 0.18 |
| | 38:6 | 6.35 | 882.5258 | -H | $C_{47}H_{79}O_{13}P$ | −2.9 | 152.9958 (PL2) | ND | B | 3.23 ± 0.12 |
| | 18:2 | 6.78 | 596.2962 | -H | $C_{27}H_{49}O_{12}P$ | −1.6 | 78.9591 (PL1), 152.9958 (PL2) | C (18:2) | A | 65.30 ± 1.49 |
| | 30:2 | 7.09 | 778.4632 | -H | $C_{39}H_{71}O_{13}P$ | 1.6 | 152.9958 (PL2) | ND | B | 3.32 ± 0.22 |
| | 18:1 | 7.55 | 598.3118 | -H | $C_{27}H_{51}O_{12}P$ | 0.0 | 152.9958 (PL2) | C (18:1) | A | 10.81 ± 0.64 |
| | 27:0 | 8.92 | 740.4476 | -H | $C_{36}H_{69}O_{13}P$ | 1.4 | 152.9958 (PL2) | ND | B | 4.64 ± 0.24 |
| | 30:0 | 9.14 | 782.4945 | -H | $C_{39}H_{75}O_{13}P$ | 0.9 | 152.9958 (PL2) | ND | B | 4.00 ± 0.22 |
| PS/ESI- | | | | | | | | | | |
| | 18:4 | 4.22 | 517.2441 | -H | $C_{24}H_{40}NO_9P$ | −1.8 | ND | ND | C | 3.10 ± 0.21 |
| | 33:2 | 6.35 | 745.4894 | -H | $C_{39}H_{72}NO_{10}P$ | 3.7 | 152.9958 (PL2) | C (18:2) C (15:0) | A | 3.51 ± 0.32 |
| | 32:3 | 7.06 | 729.4581 | -H | $C_{38}H_{68}NO_{10}P$ | 4.0 | 152.9958 (PL2) | ND | B | 3.84 ± 0.23 |
| | 19:0 | 8.34 | 539.3223 | -H | $C_{25}H_{50}NO_9P$ | 0.1 | 224.0330 (PS) | ND | B | 3.60 ± 0.13 |
| | 27:1 | 8.52 | 663.4111 | -H | $C_{33}H_{62}NO_{10}P$ | −0.2 | ND | ND | C | 3.22 ± 0.12 |
| | 30:2 | 8.52 | 703.4424 | -H | $C_{36}H_{66}NO_{10}P$ | −0.7 | ND | ND | C | 3.21 ± 0.09 |
| | 31:5 | 8.53 | 711.4111 | -H | $C_{37}H_{62}NO_{10}P$ | −1.3 | ND | ND | C | 3.15 ± 0.08 |
| | 40:2 | 9.25 | 843.5989 | -H | $C_{46}H_{86}NO_{10}P$ | 0.4 | ND | ND | C | 3.24 ± 0.12 |
| | 36:4 | 10.26 | 783.5050 | -H | $C_{42}H_{74}NO_{10}P$ | 0.4 | 78.9591 (PL1), 152.9958 (PL2) | C (18:2) C (18:2) | A | 13.57 ± 0.24 |
| | 43:0 | 15.88 | 877.6772 | -H | $C_{48}H_{96}NO_{10}P$ | 0.1 | ND | ND | C | 3.12 ± 0.22 |

Cite: ND represents not detected in the identification process.
A represents identified via mass error < 5 ppm, matched with characteristic fragments and fatty acid acyl chain fragments; B represents identified via mass error < 3 ppm and matched with characteristic fragments or fatty acid acyl chain fragments; C represents identified only via mass error < 3 ppm and the peak abundance < 1500.

**Table 3**
Identified PLs from *S. chinensis* oil in the positive ion mode.

| Head | Species | RT/min | Neutral mass | Adducts | Formula | Mass error(ppm) | $MS^E$-H: Feature fragments | Degree | Semi-quantitive content (μg/g) |
|---|---|---|---|---|---|---|---|---|---|
| PC/$ESI^+$ | | | | | | | | | |
| | 14:0 | 6.28 | 467.3012 | $+NH_4$ | $C_{22}H_{46}NO_7P$ | −1.4 | ND | C | 3.57 ± 0.08 |
| | 18:3 | 6.52 | 517.3168 | +Na | $C_{26}H_{48}NO_7P$ | 2.9 | ND | C | 3.52 ± 0.06 |
| | 22:5 | 6.69 | 569.3481 | $+NH_4$ | $C_{30}H_{52}NO_7P$ | −0.9 | 184.07332 (PC) | B | 3.96 ± 0.12 |
| | 26:1 | 6.97 | 647.4526 | +Na | $C_{34}H_{66}NO_8P$ | −3.1 | 184.07332 (PC) | B | 3.70 ± 0.09 |
| | 18:2 | 7.20 | 519.3325 | +Na | $C_{26}H_{50}NO_7P$ | 0.7 | 184.07332 (PC) | B | 9.02 ± 0.21 |
| | 18:1 | 7.73 | 521.3481 | +Na | $C_{26}H_{52}NO_7P$ | −2.1 | 184.07332 (PC) | B | 3.63 ± 0.03 |
| | 16:0 | 7.76 | 495.3325 | +Na | $C_{24}H_{50}NO_7P$ | 3.2 | 184.07332 (PC) | B | 4.51 ± 0.04 |
| | 36:5 | 8.83 | 779.5465 | +Na | $C_{44}H_{78}NO_8P$ | −1.8 | ND | C | 3.62 ± 0.04 |
| | 34:2 | 9.83 | 757.5622 | +Na | $C_{42}H_{80}NO_8P$ | −2.2 | 184.07332 (PC) | A | 3.71 ± 0.05 |
| | 36:4 | 10.00 | 781.5622 | +Na | $C_{44}H_{80}NO_8P$ | −2.1 | 184.07332 (PC) | A | 39.68 ± 0.68 |
| | 33:4 | 10.05 | 739.5152 | +Na | $C_{41}H_{74}NO_8P$ | −1.7 | 184.07332 (PC) | A | 15.54 ± 0.21 |
| | 36:3 | 10.19 | 783.5778 | +Na | $C_{44}H_{82}NO_8P$ | −2.3 | 184.07332 (PC) | A | 19.52 ± 0.21 |
| | 36:2 | 10.44 | 785.5935 | +Na | $C_{44}H_{84}NO_8P$ | 0.2 | 184.07332 (PC) | A | 7.15 ± 0.18 |
| | 31:1 | 10.50 | 717.5309 | +Na | $C_{39}H_{76}NO_8P$ | −1.9 | ND | C | 3.64 ± 0.05 |
| PA/$ESI^+$ | | | | | | | | | |
| | 24:0 | 4.64 | 536.3478 | +Na | $C_{27}H_{53}O_8P$ | 1.1 | ND | C | 3.61 ± 0.04 |
| | 20:4 | 6.61 | 458.2433 | $+NH_4$ | $C_{23}H_{39}O_7P$ | 2.1 | ND | C | 3.69 ± 0.04 |
| | 18:2 | 7.41 | 434.2433 | +Na | $C_{21}H_{39}O_7P$ | 0.1 | ND | C | 3.69 ± 0.05 |
| | 18:1 | 7.62 | 436.2590 | $+NH_4$ | $C_{21}H_{41}O_7P$ | 2.6 | ND | C | 3.50 ± 0.06 |
| | 22:2 | 9.66 | 490.3059 | $+NH_4$ | $C_{25}H_{47}O_7P$ | −2.9 | ND | C | 3.65 ± 0.05 |
| | 40:4 | 10.11 | 752.5356 | +Na | $C_{43}H_{77}O_8P$ | 0.1 | ND | C | 3.67 ± 0.03 |
| | 33:0 | 10.67 | 648.5094 | +Na | $C_{36}H_{73}O_7P$ | 2.4 | ND | C | 3.56 ± 0.03 |
| | 34:0 | 10.73 | 676.5043 | +Na | $C_{37}H_{73}O_8P$ | 0.3 | ND | C | 3.56 ± 0.04 |
| | 40:1 | 11.26 | 758.5826 | +Na | $C_{43}H_{83}O_8P$ | 2.4 | ND | C | 3.70 ± 0.04 |
| | 40:0 | 11.71 | 760.5982 | +Na | $C_{43}H_{85}O_8P$ | −0.1 | ND | C | 3.62 ± 0.08 |
| PE/$ESI^+$ | | | | | | | | | |
| | 17:0 | 6.28 | 467.3012 | $+NH_4$ | $C_{22}H_{46}NO_7P$ | −1.4 | ND | C | 3.57 ± 0.07 |
| | 18:2 | 7.08 | 477.2855 | +Na | $C_{23}H_{44}NO_7P$ | 1.7 | ND | C | 3.67 ± 0.06 |
| | 32:3 | 7.40 | 685.4683 | $+NH_4$ | $C_{37}H_{68}NO_8P$ | −3.0 | ND | C | 3.61 ± 0.08 |
| | 31:4 | 10.50 | 717.5309 | +Na | $C_{39}H_{76}NO_8P$ | −1.9 | ND | C | 3.64 ± 0.11 |
| PI/$ESI^+$ | | | | | | | | | |
| | 20:5 | 5.48 | 618.2805 | +Na | $C_{29}H_{47}O_{12}P$ | −1.2 | ND | C | 3.57 ± 0.08 |
| | 18:3 | 6.09 | 594.2805 | +Na | $C_{27}H_{47}O_{12}P$ | −0.7 | ND | C | 3.53 ± 0.04 |
| | 20:2 | 6.40 | 624.3275 | +Na | $C_{29}H_{53}O_{12}P$ | 0.9 | ND | C | 3.47 ± 0.03 |
| | 20:0 | 8.00 | 628.3588 | +Na | $C_{29}H_{57}O_{12}P$ | −1.9 | ND | C | 3.52 ± 0.02 |
| | 31:3 | 9.32 | 790.4632 | +Na | $C_{40}H_{71}O_{13}P$ | −2.0 | ND | C | 3.64 ± 0.04 |
| | 40:6 | 9.87 | 910.5571 | $+NH_4$ | $C_{49}H_{83}O_{13}P$ | 0.7 | ND | C | 3.60 ± 0.09 |
| | 40:4 | 9.97 | 914.5884 | $+NH_4$ | $C_{49}H_{87}O_{13}P$ | 0.8 | ND | C | 3.60 ± 0.12 |
| PS/$ESI^-$ | | | | | | | | | |
| | 18:0 | 8.32 | 525.3067 | +Na | $C_{24}H_{48}NO_9P$ | 2.1 | ND | C | 3.51 ± 0.03 |
| | 39:4 | 10.49 | 825.5520 | +Na | $C_{45}H_{80}NO_{10}P$ | 0.8 | ND | C | 3.49 ± 0.04 |
| | 36:7 | 10.61 | 777.4581 | $+NH_4$ | $C_{42}H_{68}NO_{10}P$ | −1.5 | ND | C | 3.49 ± 0.04 |
| | 36:0 | 11.88 | 791.5676 | $+NH_4$ | $C_{42}H_{82}NO_{10}P$ | −0.1 | ND | C | 3.55 ± 0.05 |

Cite: ND represents not detected in the identification process.
A represents identified via mass error < 5 ppm, matched with characteristic fragments and fatty acid acyl chain fragments; B represents identified via mass error < 3 ppm and matched with characteristic fragments or fatty acid acyl chain fragments; C represents identified only via mass error < 3 ppm and the peak abundance < 1500.

(Nonlinear Dynamics, Newcastle, U.K) and QI analysis software (Nonlinear Dynamics, Newcastle, U.K).

### 2.4. Identification of PLs in S. chinensis oil

For the identification of PLs in *S. chinensis* oil by UPLC-Q/TOF-$MS^E$. The MS data of PLs were collected simultaneously under positive and negative ion mode. All of the data were acquired in a single analysis process. The concrete analysis steps were as follows:

First, we created and collected the component library based on LIPID MAPS and imported it into the software (UNIFI 1.8.1). Second, we added the detailed adducted fatty acids ions and feature fragment information of each PL classes into analysis method. Third, we processed the sample data to obtain the MS spectral information of PLs. Finally, we analyzed the PL components in *S. chinensis* oil based on the MS spectra ($MS^E$-L and $MS^E$-H). It is worth mentioning that the adduct ion mode of fatty acid was based on the PLs fragmentation patterns and all the typical feature fragments information of PL classes were listed in Table 1.

As shown in Table 1, in the positive mode, the feature fragments of phosphatidylcholines (PCs), phosphatidylethanolamines (PEs), phosphatidylglycerols (PGs), phosphatidylserines (PSs) and phosphatidylinositols (PIs) were *m/z* 184.0732, 142.0269, 88.0399 and 226.0481, and 261.0735, respectively. In the negative ion mode, except for PCs, the rest of PLs owned various feature fragments (Murphy and Axelsen, 2011; Ali et al., 2017). For phosphatidic acid (PAs), PEs, PSs and PGs, they shared the common feature fragments of *m/z* 78.9585 and 171.0059. Besides, the *m/z* 152.9953 was the common fragments of PAs, PSs, PGs and PIs. Furthermore, the typical fragments of PEs were *m/z* 140.0113. The *m/z* 224.0324 was assigned to the class of PSs. PGs owned the fragment of *m/z* 227.0321, while *m/z* 259.0219, 241.0113 and 299.0532 belonged to PIs (Murphy and Axelsen, 2011; Song et al., 2017). The main adduct ions of PLs in negative and positive ion mode were $[M-H]^-$, $[M+Na]^+$ and $[M+NH_4]^+$, respectively.

### 2.5. Identification of phytosterols in S. chinensis oil

The identification process of phytosterols in *S. chinensis* oil by UPLC-Q/TOF-$MS^E$ was as follows: First, a components library was created using LIPID MAPS, including their name, formula and structure. Next,

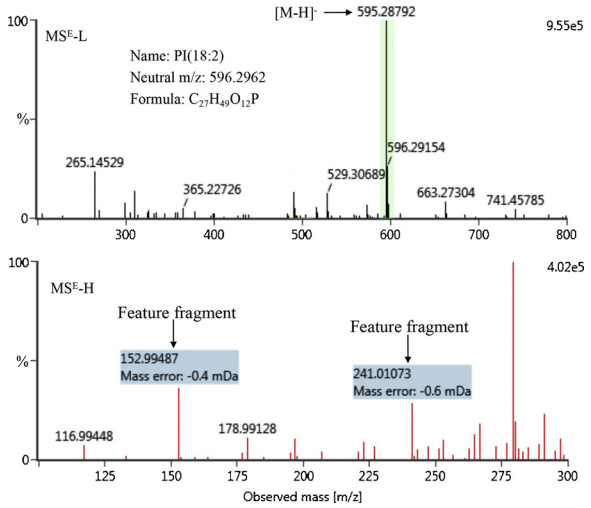



**Fig. 1.** Typical MS spectrum of PI (18:2) in *S. chinensis* oil under negative ion mode.

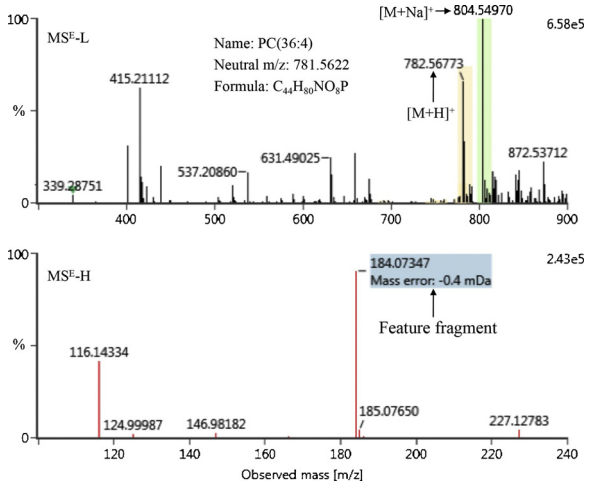



**Fig. 2.** Typical MS spectrum of PC (36:4) in *S. chinensis* oil under positive ion mode.

the scan data of $MS^E$-L and $MS^E$-H were collected in the positive ion mode. Then, the $MS^E$-L information was compared with mother ions and $MS^E$-H information with fragment ions. Finally, the phytosterols were deduced according to their MS information and previous reports. (Alberici et al., 2016; Nestola and Schmidt, 2016; Gachumi and El-Aneed, 2017).

### 2.6. Quantitative analysis of PLs and phytosterols

All the identified PLs and phytosterols in *S. chinensis* oil were quantified by external standard method. The standard of PC (36:4) was accurately weighed and dissolved in methanol, giving the mother solution of 1.00 mg/mL. Then, the mother solution was diluted into 20.00 μg/mL by methanol. Finally, four-fold step dilution was conducted for five times. The concentration of the standard solution ranged from $1.95 \times 10^{-2}$ to 20.00 μg/mL. The concentration of stigmasterol ranged from 1.00 μ g/mL to 0.10 mg/mL. The calibration equation was established according to the chromatographic ion response value of standard and its corresponding concentrations.

**Table 4**
The composition of phytosterols in *S. chinensis* oil determined by UPLC-Q/TOF-$MS^E$.

| Code | Compound name | RT (min) | Neutral Mass (Da) | Mass error (ppm) | Molecular formula | $MS^E$-L $[M+A]^+$ | $MS^E$-H[m/z] and relative abundance (%) | Semi-quantitive content (μg/g) |
|---|---|---|---|---|---|---|---|---|
| 1 | Secosterol-B | 3.73 | 418.3447 | 3.8 | $C_{27}H_{46}O_3$ | $-H_2O+H$ | [401](100), [337](80), [254](57), [175](52), [168](7) | 21.22 ± 1.21 |
| 2 | Corbisterol | 3.89 | 410.3549 | 2.3 | $C_{29}H_{46}O$ | $+H$ | [161](100), [158](86), [143](36), [130](24), [91](18) | 5.05 ± 0.42 |
| 3 | Brassicasterol | 3.97 | 398.3549 | 3.6 | $C_{28}H_{46}O$ | $+H$ | [143](100), [323](58), [337](36) | 21.88 ± 1.02 |
| 4 | Stigmasterol | 4.33 | 412.3705 | −0.2 | $C_{29}H_{48}O$ | $+H$ | [128](100), [177](67), [341](34), [105](22), [147](13) | 5.17 ± 0.32 |
| 5 | Ergosterol | 6.00 | 396.3392 | −1.5 | $C_{28}H_{44}O$ | $+H$ | [336](100), [215](91), [157](80), [161](66), [203](39) | 7.51 ± 0.23 |
| 6 | Campesterol | 6.39 | 400.3705 | 2.7 | $C_{28}H_{48}O$ | $+H$ | [161](100), [131](32), [157](29), [109](10), [201](9) | 21.57 ± 0.98 |
| 7 | Herbasterol | 6.81 | 468.3451 | 1.2 | $C_{27}H_{48}O_6$ | $+Na, +H$ | [55](100), [83](89) | 8.74 ± 0.43 |
| 8 | Lanosterol | 6.78 | 426.3862 | 1.8 | $C_{30}H_{50}O$ | $-H_2O+H$ | [145](100), [203](81), [119](71), [107](56), [409](40) | 7.14 ± 0.34 |
| 9 | Conicasterol C | 7.09 | 458.3760 | 0.8 | $C_{30}H_{50}O_3$ | $-H_2O+H$ | [203](100), [423](56), [337](43), [159](25), [121](18) | 22.02 ± 0.98 |
| 10 | Hipposterol | 8.49 | 420.3604 | −2.0 | $C_{27}H_{48}O_3$ | $+Na, +H$ | [145](100), [119](86), [245](59), [131](47), [209](26) | 4.85 ± 0.32 |
| 11 | Penasterol | 8.78 | 456.3604 | −1.7 | $C_{30}H_{48}O_3$ | $+Na$ | [259](100), [201](72), [109](38), [191](16) | 7.32 ± 0.32 |
| 12 | Theonellasterol D | 9.01 | 472.3917 | 2.9 | $C_{31}H_{52}O_3$ | $-H_2O+H$ | [203](100), [161](66), [293](50), [189](27), [217](15) | 3.84 ± 0.12 |
| 13 | Conicasterol D | 9.91 | 444.3604 | 0.9 | $C_{29}H_{48}O_3$ | $+H$ | [415](100), [317](78), [218](10) | 4.22 ± 0.22 |

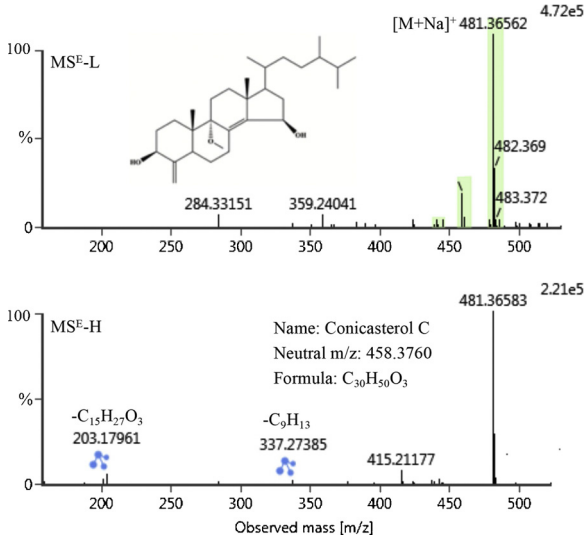



**Fig. 3.** Typical MS spectrum of conicasterol C in *S. chinensis* oil under positive ion mode.

## 3. Results and discussion

### 3.1. Molecular species of PLs in S. chinensis oil

The composition of PLs might be used to evaluate the biological activity of *S. chinensis* oil. And the profile of PLs in *S. chinensis* oil was separated efficiently by artificial adjustment of the elution conditions of $MS^E$ detector. The total identified PLs in *S. chinensis* oil under negative and positive ion mode were 49 and 39, respectively (Tables 2 and 3). They included the PLs class of PAs, PEs, PGs, PIs, PSs and PCs. All the identified components were assigned with different capital letters A, B and C that represents the accuracy degree of identification. The capital letter "A" represented the maximum probability of accuracy. Capital letter "B" represented more than half of the possibility in identification accuracy. Capital letter "C" represented the possible components with lower probability. The PLs identified in *S. chinensis* oil were classified based on the above-mentioned principles.

In order to make a more intuitive explanation of the identification of PLs, the MS spectra of the most abundant species were selected as a model for interpretation. In the negative ion mode, PI (18:2) was the most abundant component in *S. chinensis* oil. The MS spectrum of PI (18:2) was shown in Fig. 1. From the $MS^E$-L spectrum, we observed that the formula of PI (18:2) was $C_{27}H_{49}O_{12}P$, and the neutral *m/z* was 596.2962. Its adduct ion was $[M-H]^-$, with the *m/z* 595.2879. The $MS^E$-H has shown that the feature fragments of PI (18:2) were *m/z* 152.9948 and 241.0107, respectively. Besides, the matched acyl chain of PI (18:2) was C (18:2). Thus, the substance was assigned to PI (18:2) and belonged to the capital letter "A".

Similarly, in positive ion mode, the most abundant component was PC (36:4) in *S. chinensis* oil. The MS spectrum of PC (36:4) was shown in Fig. 2. The neutral *m/z* of PC (36:4) was 781.5622, with a formula of $C_{44}H_{80}NO_8P$. From the $MS^E$-L spectrum, it can be seen that the adduct ion of PCs were $[M+H]^+$ (*m/z* 782.5677) and $[M+Na]^+$ (*m/z* 804.5497). The $MS^E$-H spectrum indicated that its feature fragment was *m/z* 184.0735, conformed to the class of PC. In addition, the matched acyl chains of PC (36:4) were C (18:2) and C (18:2), respectively. Therefore, the component was identified as PC (36:4) and assigned with the capital letter "A". All the PL species in *S. chinensis* oil were tentatively identified according to the regulation as mentioned earlier.

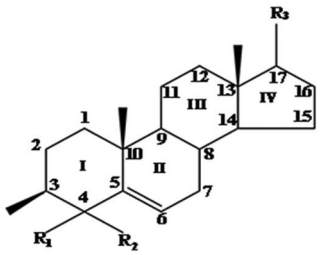
Specifically, the PLs identified in *S. chinensis* oil under negative ion mode were shown in Table 2. As shown in Table 2, except for the class of PCs, all the other types of PLs were detected in negative ion mode. The species of each PL class were eight PAs, ten PEs, nine PGs, 12 PIs and ten PSs. The total number of capital letter "A" components were 12, including three PEs, three PGs, four PIs and two PSs. In addition, the fatty acid of C (16:0) and C (18:2) were the most abundant common acyl chain in all kinds of PLs. The composition of PLs in *S. chinensis* oil under positive ion mode was shown in Table 3. The identified molecular species of PAs, PCs, PEs, PIs, PSs in *S. chinensis* oil were ten, 14, four, seven, and four, respectively. And the total components assigned to the capital letter "A" was five, corresponding to PC (34:2), PC (36:4), PC (33:4), PC (36:3) and PC (36:2). The rest species of PLs were only classified as capital letter "B" or "C" in positive ion mode. The percentage of the identified PLs in *S. chinensis* oil belonging to the capital letter "C" was 74.36%. The reason might be that the PLs species owned a weaker MS ion response in positive ion mode.

### 3.2. Content of PLs in S. chinensis oil

The standard component of PC (36:4) has been detected under negative and positive ion mode and used for quantification analysis. The equation in negative and positive were $y = 2.89 \times 10^4 x - 7987.72$, $R^2 = 0.9992$, and $y = 4.12 \times 10^4 x - 1.37 \times 10^4$, $R^2 = 0.9998$, respectively. Each of them showed good linearity and an excellent coefficient. The limit of detection (LOD) was determined based on the signal to noise ratio of three. The limit of quantification (LOQ) was calculated as the lowest injection concentration of the detected components. The LOD of PLs in negative and positive were $5.12 \times 10^{-3}$ and $12.4 \times 10^{-3}$ respectively. The LOQ of PLs in both ion mode was $1.95 \times 10^{-2}$ μg/mL. The RSDs of quantification of PLs in negative mode ranged from 2.54 to 8.03%, and the RSDs of quantification of PLs in positive mode ranged from 0.54 to 3.12%. The response of PC (36:4) in positive mode was stronger than in negative ion mode, which might be due to the class of PCs owned a stronger response in positive ion mode than negative ion mode.

The composition of PLs in *S. chinensis* oil under negative ion mode was discussed in Table 2. According to the quantification analysis, the most abundant PL class in negative ion mode was PEs, accounting for 177.68 ± 4.02 μg/g, composing 43.92% of the total identified components. The class of PAs owned the lowest content of 31.67 ± 0.84 μg/g, composing 7.83% of the total identified components. The species of PLs assigned with the capital letter "A" in *S. chinensis* oil were described as follows. For the class of PEs, they were PE (18:2), PE (16:0), PE (36:4) and PE (34:2), respectively. Their acyl chains were C (18:2) and C (16:0). PE (18:2) was the most abundant substance, the content being 61.44 ± 2.64 μg/g. Similarly, the species of PGs were PG (18:2), PG (16:0) and PG (41:0), with the acyl chain of C (18:2), C (16:0), C (20:0) and C (21:0), respectively. PG (16:0) was the most abundant component, with the content of 6.28 ± 0.42 μg/g. PI (35:6), PI (36:6), PI (31:2), PI (18:2) and PI (18:1) were the species of PIs marked with capital letter "A". The fatty acyl chains of those PI species were C (18:2), C (18:4), C (14:0), C (17:2) and C (18:1), respectively. PI (18:2) possessed the highest content of 65.30 ± 2.88 μg/g. Besides, the marked "A" components of PSs class were PS (33:2) and PS (36:4), and PS (36: 4) reached the highest content of 13.57 ± 0.24 μg/g. Their typical acyl chains were C (18:2) and C (15:0). In a word, it can be drawn that *S. chinensis* oil contained abundant and diverse kinds of PLs, and C (18:2) and C (16:0) were the most abundant fatty acyl chains of different PLs.

The specific information of PLs in *S. chinensis* oil under positive ion mode was shown in Table 3. The separation efficiency of PLs in positive ion mode was weaker than in the negative ion mode. Except for the PCs, the other class of PL species were lacked sufficient evidence to make a full identification. The percentage of PAs, PCs, PEs, PIs and PSs were 18.28, 57.21, 6.65, 11.43 and 6.44%, respectively. It is worth noticing that the total content of confirmed PC species was 85.61 ± 2.12 μg/g, namely PC (34:2), PC (36:4), PC (33:4), PC (36:3) and PC (36:2). The content of the rest components owned a smaller fluctuation, which was less than 4.00 μg/g. Accordingly, we could draw the conclusion that the

| General formula | Name | $R_1$ | $R_2$ | $R_3$ | DB-N | DB-P |
|---|---|---|---|---|---|---|
|  | Corbisterol | - | - | $-C_9H_{18}$ | 3 | 5,6; 7,8; $R_3$ |
| | Brassicasterol | - | - | $-C_8H_{16}$ | 2 | 5,6; $R_3$ |
| | Stigmasterol | - | - | $-C_9H_{18}$ | 2 | 5,6; $R_3$ |
| | Ergosterol | - | - | $-C_8H_{16}$ | 3 | 5,6; $R_3$ |
| | Campesterol | - | - | $-C_8H_{18}$ | 1 | 5,6; |
| | Lanosterol | $-CH_3$ | $-CH_3$ | $-C_7H_{14}$ | 2 | 8,9; $R_3$ |

Cite: DB-N represents for double-bond numbers. DB-P represents for double-bond positions

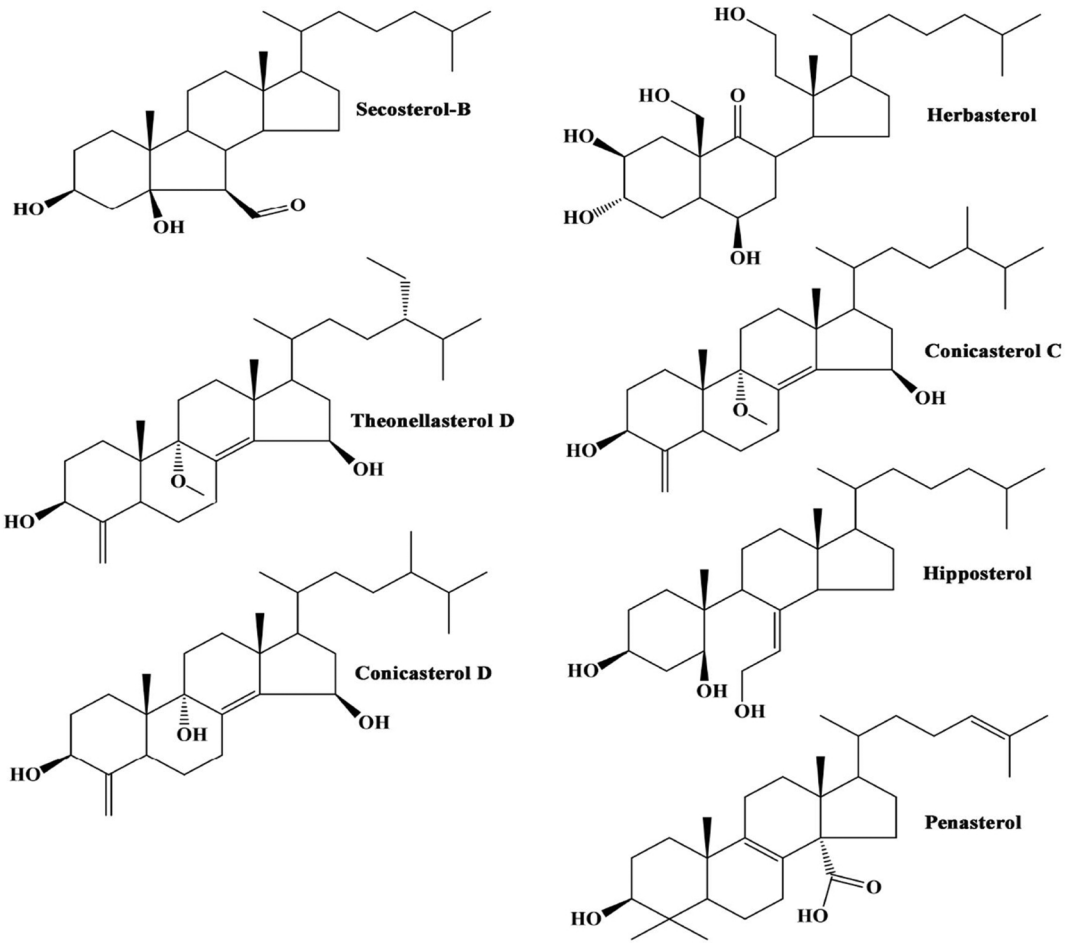



**Fig. 4.** Chemical structure of phytosterols components identified in *S. chinensis* oil.

class of PCs should be detected in the positive ion mode and the rest class of PLs should be analyzed in the negative ion mode. The composition of PLs in *S. chinensis* oil might be an excellent indication for its further application and quality evaluation.

### 3.3. Phytosterols composition in the S. chinensis oil by UPLC-Q/TOF-MS

Phytosterols are cholesterol-like molecules found at high concentrations in vegetable oils and contribute to inhibiting the absorption of intestinal cholesterol (Hu et al., 2018). In this study, the composition of phytosterols in *S. chinensis* oil was analyzed by UPLC coupled with the $MS^E$ detector. They were determined in the positive ion mode. It is worth noticing that the identification of phytosterols seems weak without verification of standards. There are phytosterol isomers that cannot be distinguished only by their exact mass. Except for the standard component of stigmasterol, the rest of them were tentatively identified. The identified components were listed in Table 4. As shown in Table 4, there were 13 kinds of phytosterols detected in the *S. chinensis* oil. The main adduct ions of phytosterols in *S. chinensis* oil were $[M\text{-}H_2O+H]^+$, $[M+Na]^+$, and $[M+H]^+$. The MS spectrum of the most abundant component conicasterol C has been shown in Fig. 3. Its adduct ion was $[M+Na]^+$, *m/z* 481.3656. The formula of conicasterol C was $C_{30}H_{50}O_3$, with the typical fragments of $-C_{15}H_{27}O_3$ and $-C_9H_{13}$. Therefore, the substance was identified as conicasterol C based on the

detailed information of $MS^E$-L and $MS^E$-H. All the phytosterols in *S. chinensis* oil were identified by the principle as mentioned earlier.

The regression equation of standard component stigmasterol was $y = 23.447\ x + 276.82$, $R^2 = 0.9861$. The LOD and the LOQ of phytosterols were 0.02 and 1.00 μg/mL, respectively. The RSDs of all the identified phytosterols ranged from 3.12 to 8.32%. The top four content of phytosterols in *S. chinensis* oil were conicasterol C, brassicasterol, campesterol, and secosterol-B, respectively. Their corresponding content were 22.02 ± 0.98, 21.88 ± 1.02, 21.57 ± 0.98 and 21.22 ± 1.21 μg/g, respectively. The rest of phytosterols in *S. chinensis* oil had the content less than 10.00 μg/g. In order to make a comprehensive understanding of phytosterols in *S. chinensis* oil, the specific information of each component has been explored thoroughly in the library of LIPID MAPS and published papers. And the chemical structure of each phytosterol in *S. chinensis* oil was shown in Fig. 4. As shown, it is easy to draw that the main difference among phytosterols identified in oils were the double-bond quantity and their positions. Based on the structure of identified phytosterols in *S. chinensis* oil, they can be categorized into two categories. One group shared similar structures and was usually detected in the edible oils, the other group possessed irregular structures and was rarely detected in oils.

The group of components with similar structures included corbisterol, brassicasterol, stigmasterol, ergosterol, campesterol and lanosterol. They have been widely identified in various vegetable oils (Li et al., 2011; Vrbková et al., 2014). Compared with common vegetable oils, the composition of phytosterol in *S. chinensis* oil was slightly different. For example, sitosterol was not detected in the *S. chinensis* oil and the content of stigmasterol was lower. The reason might be that the *S. chinensis* is an herbal plant, and its chemical components are different from ordinary plants.

With regard to the group of irregular structures, many of them were detected in the herbal plants and marine creatures, including herbasterol, conicasterol C, hipposterol, penasterol, theonellasterol D and conicasterol D (De Marino et al., 2011; Lyakhova et al., 2015). And the secosterol-B which belonged to the saturated phytosterol has been detected in human atherosclerotic plaques and tissue samples of brains. It was a derivative of cholesterol, playing an important role in inflammation-related diseases (Okada et al., 2018). The top three content of this group of phytosterols in *S. chinensis* oil was conicasterol C, herbasterol and penasterol. The content of herbasterol and penasterol in *S. chinensis* oil were 8.74 ± 0.83 and 7.32 ± 0.32 μg/g, respectively. It is reported that herbasterol and penasterol were detected in the extraction of marine sponges and Chinese medicinal herb plants. They possessed the function of anti-leukemia activity and inhibiting of histamine released from rat mast cells (Capon and Faulkner, 1985; Lu et al., 2007; Lyakhova et al., 2015). The components of conicasterol C, theonellasterol D and conicasterol D were separated from several sponges of the genus *Theonella*. They were the symbol feature of this species, and these kinds of phytosterols could be potentially applied in the treatment of liver disorders (De Marino et al., 2011). The last component hipposterol was isolated from the bamboo coral *Isis hippuris,* possessing the bioactivity of anti-human cytomegalovirus (Chen et al., 2011). All the above-mentioned statements indicated that *S. chinensis* oil contains various kinds of phytosterols, and most of them are highly pharmacologically active.

## 4. Conclusions

A systematic analysis of PLs and phytosterols in *S. chinensis* oil has been performed in UPLC-Q/TOF-$MS^E$ operation. The combination of multistage collision and high mass resolution in Vion-IMS-Q-ToF system provided feature fragment and acyl chain information of PLs under both ion modes. A total of 49 and 39 PL molecular species were identified in negative and positive ion mode from *S. chinensis* oil, and the components were quantified by external standard method. Thirteen kinds of phytosterols were identified in *S. chinensis* oil under positive ion mode, and their specific information has also been provided. In conclusion, the composition of PLs and phytosterols in *S. chinensis* oil were investigated in detail for the first time, which could establish a foundation for further study on nutritive function and product application of *S. chinensis* oil.

## Conflict of interest

The authors declare that there is no conflict of interests regarding this paper publication.

## Acknowledgments

This work was supported by the National Natural Science Foundation of China (Nos. 31671958 and 31471668), Shanghai Jiao Tong University Agri-X Fund (No. Agri-X2015007).

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